Chromosome Numbers for Eight Species in Five Genera of Menispermaceae

Heng-Chang WANG, Ai-Ping MENG, Jian-Qiang LI* and Zi-Can HE

Wuhan Botanical Garden, Wuhan Institute of Botany, the Chinese Academy of Sciences, Wuhan, 430074 CHINA *E-mail to the author for correspondence: jianqiangli@hotmail.com

(Received on September 13, 2003)

Chromosome numbers of 8 species in 5 genera of Menispermaceae from China are reported: $Tinospora\ sagittata\ (Oliv.)\ Gagnep.\ 2n=4x=52,\ and\ T.\ hainanensis\ Luo\ \&\ Z.$ X. Li 2n=26; $Diploclisia\ glaucescens\ (Bl.)\ Diels\ 2n=26$; $Stephania\ yunnanensis\ Luo\ 2n=26$, and S. $dielsiana\ Wu\ 2n=26$; $Cocculus\ orbiculatus\ (L.)\ DC.\ 2n=4x=52,\ and\ C.\ laurifolius\ DC.\ 2n=26$; $Menispermum\ dahuricum\ DC.\ 2n=4x=52$. The chromosome numbers of the first five species are reported here for the first time. All eight species studied have a basic chromosome number x=13, and chromosome numbers are generally consistent with previous reports for the same species and genera.

Key words: Chromosome number, Cocculus, Diploclisia, Menispermaceae, Tinospora.

Introduction

Menispermaceae, consisting of five tribes, 71 genera and about 450 species, is a pantropical family usually confined to the lowlands. All Menispermaceae are typically unisexual and dioecious. Most are lianas and many have pharmaceutical properties and are poisonous (Kessler 1993).

Nineteen genera and 78 species of Menispermaceae are recognized in China. They are mainly distributed in the southern regions of the Yangtze River, and especially in South China and Southwest China (Luo 1996).

Chromosome numbers can sometimes provide information on the relationship of genera or even of families (Forman 1986). Raven (1975) used chromosome information to postulate a relationship between Menispermaceae and Lardizabalaceae. Forman (1986) proposed *Sabia* Colebr. (Sabiaceae) to be a near ally of Menispermaceae, based on chromosome data.

Although cytological studies in Menispermaceae have been published by various authors (Darlington and Wylie 1955, Fedorov 1974, Goldblatt 1984, 1985, Hong 1990, etc.), chromosome counts have been made in only a few species in each of seven of the 25 Malesian genera, viz. *Tiliacora*, *Tinospora*, *Anamirta*, *Cocculus*, *Cissampelos*, *Cyclea* and *Stephania* (Forman 1986).

The Chinese species of Menispermaceae, however, are poorly treated in most standard references. Chromosome counts for only two species from China, $Cocculus\ trilobus\ (=C.\ orbiculatus)$ reported by Nakajima (1937) as 2n=50 and $Menispermum\ dauricum\ from$ North China observed by Langlet (1928) as 2n=52-54, have been reported. In this paper, eight species in five genera of Menispermaceae from China were studied to determine chromosome number and other cytological characteristics.

Material and Methods

The plants investigated were collected from the original locality, but now they have been cultivated in South China Botanical Garden, the Chinese Academy of Sciences (CAS) and Wuhan Botanical Garden, CAS (Table 1). Voucher specimens were deposited in the herbarium of the Wuhan Institute of Botany, Chinese Academy of Sciences (HIB).

For observations of somatic chromosomes, growing shoot apices were pretreated in a saturated solution of p-Dichlorobenzene at about 18–20°C for 3.5–4 hours, fixed in Carnoy I (glacial acetic acid: absolute ethanol = 1:3) at the same temperature for 24 hours, and then transferred to 70 % alcohol. The shoot apices were hydrolyzed in a 1:1 mixture of 1 N hydrochloric acid (HCL) and 45% acetic acid at 60 °C for 6–8 minutes, and then stained in carbol fuchsin for 30 minutes and squashed in 45 % acetic acid. More than 10 metaphase cells were examined to determine the chromosome number of each species.

Results and discussion

Chromosome numbers of the species studied are given in Table 1.

1. Tinospora Miers — According to Kessler (1993), there are about 32 species in Tinospora. Seven are in tropical Africa, two in Madagascar, and twenty-three are from Asia to Australia and the Pacific. Six species and two varieties have been recorded from China (Luo 1996).

Two species had been examined previously. *Tinospora tomentosa* Miers was reported as 2n = 26 (Chattopadhyay and Sharma 1989). Joshi (1934) and Nanda (1962) examined *T. cordifolia* Miers. from India and reported it to be 2n = 24, while Abraham (1942), Bir et al. (1987), Mathew (1958), Sharma and Bhattacharyya (1955), Sharma and Sharma (1957), and Sarkar et al. (1980) reported it to be 2n = 26.

Two species, T. sagittata (Oliv.) Gagnep. and T. hainanensis Lo & Z. X. Li are reported here for the first time. All five populations of T. sagittata were found to be tetraploids with 2n = 4x = 52 (Fig. 1, 1),

| Table 1. Chromosome counts and source of materials studied in the family Menispermace | eae |
|---|-----|
|---|-----|

| Species | Locality | Sex | Voucher* | Chromosome number (2n) | Previous reports (2n) |
|-----------------------------------|--|--------|----------------------|------------------------|---|
| Tinospora sagittata | Guangdong and Hainan (SCBG)** | male | 03-008 | 52 | none |
| T. hainanensis | Lingao, Wenchang County, Hainan (SCBG) | male | 03-009 | 26 | none |
| Cocculus laurifolius | Mt. Dinghu, Guangdong (SCBG) | male | 03-004 | 26 | 26 (Bowden 1945, Gill et al. 1981, Sandhu and Mann 1988) |
| C. orbiculatus (= C. tribolus) | Lingao, Wenchang and Wanlin County, Hainan (SCBG) | male | 03-001 | 52 | 52 (Bowden 1945) 50 (Nakajima 1937, from Japan and China) |
| Diploclisia glauces- cens | Hainan (SCBG) | female | 03-006 | 26 | none |
| Menispermum dahu- ricum | Shennongjia, Hubei (WHBG)*** | male | 03-010 | 52 | 52-54 (Langlet 1928, from N. China) |
| Stephania dielsiana | Hebao Island, Guangdong (SCBG) | female | 03-002 | 26 | none |
| S. yunnanensis | Yunnan and Guangxi (SCBG) | male | H. C. Wang 03-007 | 26 | none |

^{*}Collection number of H. C. Wang.

^{**}SCBG = South China Botanical Garden, the Chinese Academy of Sciences (CAS).

^{***}WHBG = Wuhan Botanical Garden, CAS.

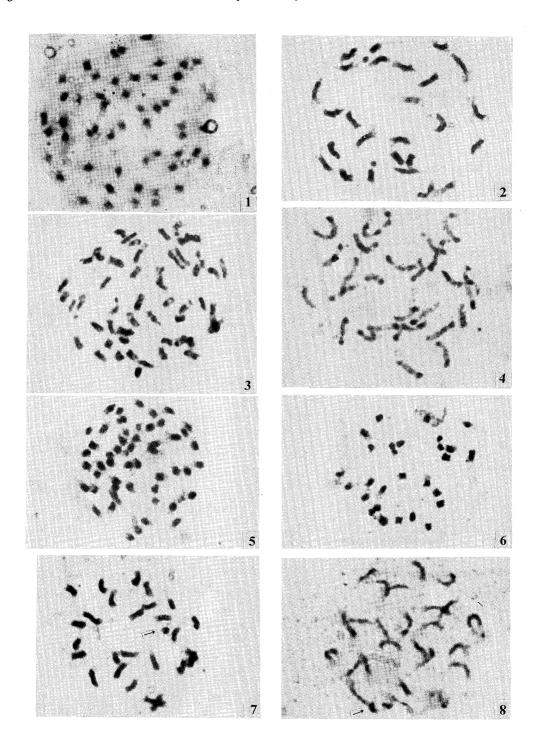


Fig. 1. Chromosomes of eight species of Menispermaceae. 1. Tinospora sagittata (Oliv.) Gagnep. 2. T. hainanensis Lo & Z. X. Li. 3. Cocculus orbiculatus (L.) DC. 4. C. laurifolius DC. 5. Menispermum dahuricum DC. 6. Diploclisia glaucescens (Blume) Diels. 7. Stephania yunnanensis Luo (Arrow indicates B-chromosome). 8. S. dielsiana Wu (Arrow indicates B-chromosome).

while six populations of T. hainanensis were 2n = 26 (Fig. 1, 2).

2. Cocculus DC. — Eight species of Cocculus are found in North and Central America, Africa, Madagascar, and eastward from the Indian subcontinent to the Philippines (Kessler 1993). Two species and one variety have been recorded from China (Luo 1996).

Chromosomes of *C. pendulus*, *C. villosus*, *C. carolinus* and *C. hirsutus* were examined by Baquar et al. (1965), Bir and Thakur (1984), Bowden (1945), Gill et al. (1981), Hagerup (1932), Joshi (1934), Lewis et al. (1962), Sarkar et al. (1973), Sharma and Bhattacharyya (1955), Sharma and Sharma (1957) and Sidhu et al. (1983).

Cocculus laurifolius was reported by Bowden (1945, materials from Himalayas and E. Asia) and Sandhu and Mann (1988) as 2n = 26, and by Gill et al. (1981) as n =13 + 0 - 3B. They are generally consistent with our report here 2n = 26 (Fig. 1, 4). While C. trilobus (Thunb.) DC. (= C. orbiculatus (L.) DC.) was reported by Bowden (1945,materials from Philippines) as 2n = 52, by Nakajima (1937, materials from Japan and China) as 2n = 50, we counted 2n = 4x = 52. We believe the basic chromosome number of the tetraploid to be x = 13 (Fig. 1, 3).

3. Menispermum L. — Two to four species of Menispermum in Eurasia and North America (Kessler 1993), and one species M. dahuricum DC. is in China (Luo 1996).

Menispermum canadense L. was reported by some authors (Langlet 1928, Lindsay 1930, Löve and Löve 1982, Tischler 1931) to be 2n = 52, and M. dauricum from North China was reported by Langlet (1928) to be 2n = 52-54 (who also reported M. canadense to be 2n = 52-54).

We examined five populations of *M*. dahuricum and determined it to be tetraploid

with 2n = 4x = 52 (Fig. 1, 5), which is consistent with Kessler's report (1993).

- 4. Diploclisia Miers Two species D. affinis (Oliv.) Diels and D. glaucescens (Blume) Diels occur in tropical Asia (Kessler 1993), and both are in China (Luo 1996). We found D. glaucescens to be 2n = 26 (first report) (Fig. 1, 6).
- 5. Stephania Lour. Stephania is somewhat confused taxonomically and the number of species is uncertain. According to Kessler (1993), there are 30 species, including five in tropical Africa and 25 from southern India to central and southern China and New Guinea. According to Luo (1996), there are about 60 species with 39 species and 1 variety in China.

The chromosome numbers of Stephania are various, with reports of 2n = 22, 24, and 26 (Darlington and Wylie 1955, Fedorov 1974, Goldblatt 1984, 1985, Hong 1990). Some Chinese species including S. elegans Hook. f. & Thomson (2n = 22), S. gracilenta Miers. (2n = 26), S. hernandifolia (Willd.) Walp. (2n = 22) and S. japonica (Thunb.) Miers. (2n = 22), were examined (Bir and Thakur 1981, 1984, Chattopadhyay and Sharma 1989, Guha 1979, Sarkar et al. 1980, Sharma and Bhattacharyya 1955, Sharma and Sharma 1957). We examined two species and found the chromosome numbers of both S. yunnanensis Luo (Fig. 1, 7) and S. dielsiana Wu (Fig. 1, 8) to be 2n = 26.

General discussion

The chromosomes of Menispermaceae are very small in size. Diploid numbers 2n = 24 and 26 are the most frequent and 2n = 22 is less common. Polyploidy is relatively rare in Menispermaceae (Thanikaimoni 1984). Although our results are not consistent with previous reports, we consider x = 13 to be one of the main basic chromosome numbers for the family.

We thank Professor Li Ze-Xian, South China Institute of Botany, the Chinese Academy of Sciences, for guidance in the field and for identification of the species. This study was financially supported by Chinese Academy of Sciences (Nos. KSCX2-SW-104, 01035108, 0105110 and 05035123).

References

- Abraham A. 1942. Chromosome number in Tinospora. Curr. Sci. 11(7): 282.
- Baquar S. R., Akhtar S. and Husain A. 1965. Meiotic chromosome numbers in some vascular plants of Indus Delta. I. Bot. Notiser 118(3): 289–298.
- Bir S. S. and Thakur. H. 1981. Chromosome number reports LXXIII. Taxon 30: 842–843.
- and _____ 1984. SOCGI plant chromosome number reports-II. J. Cytol. Genet. **19**: 114–115.
- ——, —— and Chatha G. S. 1987. Chromosomal studies in certain members of Ranunculaceae and Mensipermaceae. Proceedings of the Indian Science Congress Association **74**(3,VI): 184–185.
- Bowden W. M. 1945. A list of chromosome number in higher plants. II. Menispermaceae to Verbenaceae. Amer. J. Bot. 32(4): 191–202.
- Chattopadhyay D. and Sharma A. K. 1989. Chromosome studies and unisexuality in Menispermaceae. Nucleus **32**: 173–179.
- Darlington C. D. and Wylie A.P. 1955. Chromosome Atlas of Flowering Plants. Pp. 13–14. G. Allen London.
- Fedorov A. 1974. Chromosome Numbers of Flowering Plants. Pp. 418–419. Otto Koeltz Science Publishers, Koenigstein.
- Forman L. L. 1986. Menispermaceae. Flora Malesiana 10: 157–253.
- Gill B. S, Bir S. S. and Singhal V. K. 1981. Chromosome number reports LXXI. Taxon **30**: 513–514.
- Goldblatt P. 1984. Index to Plant Chromosome Numbers 1979–1981. Missouri Botanical Garden.
- —— 1985. Index to Plant Chromosome Numbers 1982–1983. Missouri Botanical Garden.
- Guha S. 1979. Cytological studies on *Stephania* Lour. Proc. Indian Sci. Congr. Assoc. (III, C) **66:** 81.
- Hagerup O. 1932. Über Polyploidie in Beziehungzu Klima, Ökologie und Phylogenie (Chromosomenzahlen aus Timbuktu). Hereditas **16**(1–2): 19–40.
- Hong D.Y. 1990. Plant cytotaxonomy. P. 304. Science Press, Beijing (in Chinese).

- Joshi A. C. 1934. Chromosome numbers in Menispermaceae. Nature(London) 134(3375): 29.
- Kessler P. J. A. 1993. Menispermaceae. *In*: Kubitzki K. (ed.), The Families and Genera of Vascular Plants 2: 402-418. Springer-Verlag, Heidelberg.
- Langlet O. F. J. 1928. Einige Beobachtungen über die Zytologie der Berberidazeen. Svensk Bot. Tidskr. 22(1-2): 169-184.
- Lewis W. H., Stripling H. L. and Ross R. G. 1962. Chromosome numbers for some angiosperms of the southern United States and Mexico. Rhodora. **64**(758): 147–161.
- Lindsay R. H. 1930. The chromosomes of some dioecious Angiosperms. Amer. J. Bot. 17(2): 152– 174.
- Löve A. and Löve. D. 1982. IOPB chromosome number reports LXXIV. Taxon 31: 120–126.
- Luo H. S. 1996. Menispermaceae. Flora Reipublicae Popularis Sinicae. Tomus **30** (1): 1–81. Sciences Press, Beijing (in Chinese).
- Mathew P. M. 1958. Studies on the Menispermaceae. Proc. Indian Acad. Sci., Sect. B, 47(5): 274–286.
- Nakajima G. 1937. Cytological studies on some dioecious plants. Cytologia, Fujii Jub. Vol. 282– 292.
- Nanda P. C. 1962. Chromosome number of some trees and shrubs. J. Indian Bot. Soc., 41(2): 271–277
- Raven P. H. 1975. The bases of angiosperm phylogeny: cytology. Ann. Missouri Bot. Gard. **62**: 724–764.
- Sandhu P. S. and Mann. S. K. 1988, SOCGI plant chromosome number reports–VII. J. Cytol. Genet. 23: 219–228.
- Sarkar A. K., Datta N. and Chatterjee U. 1973. Chromosome survey of certain angiosperms. II. Bull. Bot. Surv. India 15: 148.
- —, and 1980. Chromosome number reports LXVII. Taxon **29**: 360–361.
- Sharma A. K. and Bhattacharyya N. K. 1955. Cytogenetics of some members of Menispermaceae. Bull. Bot. Soc. Bengal 9(2): 159–169.
- and Sharma A. 1957. Investigations leading to a new theory of differentiation in plant cells. Genetic Iberica 9(3): 143–157.
- Sidhu M., Bir S. S. and Lata P. 1983. IOPB Chromosome number reports LXXIX. Taxon 32: 322.
- Thanikaimoni 1984. Ménispermacées: palynologie et systématique. Trav. Sect. Sci. Tech. Inst. Fr. Pondichery.
- Tischler G. 1931. Pflanzliche Chromosomen-Zahlen. Tabul. Biol. Periodicae 7: 109–226.

王 恒昌, 孟 愛平, 李 建強, 何 子灿:ツヅラフジ科 5 属 8 種の染色体数

中国産ツヅラフジ科 5 属 8 種について染色体数を報告した。そのうち、Tinospora sagittata (2n = 52)、Tinospora hainanensis (2n = 26)、Diploclisia glaucescens (2n = 26)、Stephania yunnanensis (2n = 26)、Stephania dielsiana (2n = 26) の 5 種については初めての染色体数の報告である。報告した 8 種

はいずれも x=13 の染色体基本数をもつ、既報の 3 種ではこれまでの報告に一致するが、Cocculus trilobus では 2n=52 で、Nakajima (1937) が報告した 2n=50 とは一致しなかった。しかし、この場合も染色体基本数は x=13 であると推定している。 (中国科学院武漢植物研究所武漢植物園)